

DEPARTMENT OF THE AIR FORCE 59TH MEDICAL WING (AETC) JOINT BASE SAN ANTONIO - LACKLAND TEXAS

20 APR 2016

MEMORANDUM FOR SGOZ

ATTN: CAPT MARY T PAWLAK

FROM: 59 MDW/SGVU

SUBJECT: Professional Presentation Approval

- Your paper, entitled Prevalence of Trypanosoma Cruzi Among U.S. Military Members
 Training in South Texas presented at/published to Disease in Nature, San Antonio, TX
 25-27 May 2016; Military Health System Research Symposium, Kissimmee, FL 15-18
 Aug 2016; American Society of Tropical Medicine and Hygiene, Atlanta, GA 13-17
 Nov 2016 with MDWI 41-108, and has been assigned local file #16163.
- 2. Pertinent biographic information (name of author(s), title, etc.) has been entered into our computer file. Please advise us (by phone or mail) that your presentation was given. At that time, we will need the date (month, day and year) along with the location of your presentation. It is important to update this information so that we can provide quality support for you, your department, and the Medical Center commander. This information is used to document the scholarly activities of our professional staff and students, which is an essential component of Wilford Hall Ambulatory Surgical Center (WHASC) internship and residency programs.
- 3. Please know that if you are a Graduate Health Sciences Education student and your department has told you they cannot fund your publication, the 59th Clinical Research Division may pay for your basic journal publishing charges (to include costs for tables and black and white photos). We cannot pay for reprints. If you are 59 MDW staff member, we can forward your request for funds to the designated wing POC.
- 4. Congratulations, and thank you for your efforts and time. Your contributions are vital to the medical mission. We look forward to assisting you in your future publication/presentation efforts.

hinder Steel Goodwin

LINDA STEEL-GOODWIN, Col, USAF, BSC Director, Clinical Investigations & Research Support

PROCESSING OF PROFESSI	ONAL MEDICAL R	RESEA	RCH/TE	CHNICAL	PUBLICATIO	NS/PRE	SENTATIONS
1. TO: CLINICAL RESEARCH 2. FROM: (Auth							A. PROTOCOL NUMBER:
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The Impact of Chagas Disease on Military	Training in Texas						
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Prevalence of Trypanosoma cruzi among U	J.S. Military Members	s Traini	ing in So	uth Texas			
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8. DO YOU NEED FUNDING SUPPORT FOR P		ES:	YES 🔀	ON [
9. IS THIS MATERIAL CLASSIFIED? YES	NO NO						
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11b. PUBLISHED ABSTRACT (List intend	ded journal.)						
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 EXPECTED DATE WHEN YOU WILL NEED NOTE: All publications/presentations are re 						ON TO DT	IC
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May 02, 2016							
13. 59 MDW PRIMARY POINT OF CONTACT ((Last Name, First Name,	, M.I., en	nail)			14. DUTY	PHONE/PAGER NUMBER
Pawlak, Mary, T. (mary.pawlak@us.af.mil)					671-8235	
15. AUTHORSHIP AND CO-AUTHOR(S) List in		ear in the	e manuscr	ript.			
LAST NAME, FIRST NAME AND M.I.	GRADE/RANK				FICE SYMBOL	INST	ITUTION (If not 59 MDW)
Primary/Corresponding Author Pawlak, Mary T.	O-3	559	THLS/ 55	59 MDG/ SC	OZ		
b. Hall, Sallie		59 M	IDW/ST				
c. Valtier, Sandra		59 M	iDW/ST				
d. Daniels, Candelaria C.						US A	rmy Public Health Con
e. Cropper, Thomas L.		559	THLS/ 5:	59 MDG/ SC	GOZ		
f. Webber, Bryant J.	O-3	559	THLS/ 5:	59 MDG/ SC	GOZ		
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DEPARTMENT OF THE AIR FORCE 59TH MEDICAL WING (AETC) JOINT BASE SAN ANTONIO - LACKLAND TEXAS

JUN 3 0 2014

FINAL IRB APPROVAL (FULL BOARD - MINIMAL RISK)

Approval Date: 27 May 2014

Principal Investigator: Capt Bryant Webber/SGOZ

IRB Reference Number: FWH20140074H (IRBNet 402055)

Assurance Number: FWA00001750, Expiration: 12 Aug 18 (Wilford Hall Ambulatory Surgical Center)

DOD Assurance #50007, Expiration: 8 Sep 16

Protocol Title: The Impact of Chagas Disease on Military Training in Texas

- 1. Your study, referenced above, is approved for <u>one year</u> as a <u>MINIMAL RISK</u> study by the Wilford Hall Ambulatory Surgical Center's Institutional Review Board (WHASC IRB). This study was previously Conditionally Approved and this is notification that all stipulations have been met therefore the study may begin. Additional items reviewed and approved by the WHASC IRB include:
 - ICD
 - HIPAA Authorization
 - Individual Investigator Agreement (Dr. Daniels)
 - Letter of Support (341 Training Squadron)
 - Letter of Support (343 Training Squadron)
 - Letter of Support (59 MDW Laboratory Services)
 - Letter of Support (737 Training Group)
 - Letter of Support (FADL)
 - CAMD SOP 23.3
 - CAMD SOP 3.7
 - Form A-1
 - Form A-2
 - Master Key and Enrollment Case Report Form
 - Ombudsmen Appointment Letter
 - CVs
 - CITI Training certificates
- 2. The expiration date on your new study is 27 May 2015. This is the date upon which the study is no longer approved, unless you submit a continuing review report, using the template provided by the Protocol Office which must be subsequently approved in no more than 365 days, as follows. The expiration date is based on this study's approval date. The IRB will use the expiration date as a fixed anniversary date. This means if next year's IRB approval occurs within 30 days of expiration, the expiration date will remain the same. The study will be reviewed in approximately 11 months for continuing review. Your first progress report, which is a request for continuation of the study, will be due to the Protocol Office no later than 14 April 2015. A continuing review report will be due approximately every 11 months after each approval, in order for the WHASC/IRB to approve continuation of the study for another year. You must submit all publications and presentations for prior approval by the WHASC Public Affairs and Protocol Office. Upon completion of your study, you must submit a final closeout report to the WHASC Protocol Office.

- 3. IRB approved documents are in IRBNet and can be seen in Review Details. Following IRB review/approval, it is the PI's responsibility to contact the WHASC Protocol Office at 292-4012 (DSN 554), within 30 days of notification to schedule a "New Study" start-up meeting. You will be given a study binder, the final IRB study approval letter, original date-stamped ICD, and any other IRB-approved documents needed for the study. Failure to schedule a "New Study" start-up meeting within the 30 day suspense will result in the study being suspended. It is the PI's responsibility to maintain IRB approved documents in the new study binder.
- 4. Only investigators listed below are approved to participate in the study (e.g., obtain consent and to interact with and/or collect identifiable information on research subjects):
- Capt Bryant Webber, Pl
- Sandra Valtier, PhD, Al
- Roger Bravo, RA/RC
- Lois Robinson, RA/RC
- Constance Kowat, RA/RC
- Michele Tavish, RA/RC

These are the only investigators identified by the WHASC IRB to have completed "IRB approved" investigator training. Any additions to this list must first be approved by the IRB by submitting an amendment, along with a copy of the investigator's training certificate.

- 5. Your MINIMAL RISK study will be forwarded to the Surgeon General's Research Compliance and Oversight Office (SGE-C) for information and concurrence.
- 6. The WHASC IRB must be notified immediately of any additional information, or changes to the approved protocol. All modifications to either the protocol or ICD must be reviewed and approved by the WHASC IRB prior to their inception.
- 7. You must comply with the information contained in the Certificate of Compliance.
- 8. If funds were requested for your study, you will be notified by the 59th Clinical Research Division Resource Manager (292-7924) concerning the status of the requested funds. YOU ARE NOT AUTHORIZED TO USE YOUR SECTION'S O&M FUNDS.
- 9. If you have any questions, the POC is Mrs. L. Elaine Turner at (210) 292-4012. Please include your project title and reference number in all correspondence or inquiries.

JOHN R. EKSTRAND, COL, USA, MC WHASC Authorized Institutional Official







Prevalence of Trypanosoma cruzl among U.S. Military Members Training in South Texas

Mary T. Pawlak, MD, MPH²; Sallie Hall²; Sandra Valtier, PhD²; Candelaria C. Daniels, PhD²; Thomas L. Cropper, DVM, MPVM, DACVPM²; Charla C. Tully, DO³; Bryant J. Webber, MD, MPH² 159th Medical Wing, Joint Base San Antonio-Lackland, Texas, USA; 2U.S. Army Public Health Command-Central, JBSA-Fort Sam Houston, Texas, USA; 3Defense Institute for Medical Operations, JBSA-Lackland, Texas, USA

since 2013. This cross-sectional study, funded to enroll 3,000 subjects, seeks to determine the prevalence and seroprevalence of acute and chronic Chagas disease among sub-populations considered at highest risk for the disease. who spend the majority of their time in the endemic field setting. After one year of enrollment (N=416 subjects), no positive cases have been identified. Current measures may San Antonio (JBSA), Texas, where Trypanosoma cruzi-infected triatomine insects have been collected in abundance This includes U.S. Air Force students and military instructors sufficient to prevent Chagas disease in this setting. potential issue for personnel who work and live on Joint Base human Chagas disease is an although further enrollment is needed.



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Background

39 reported cases in Texas, USA, in 2013-14, 12 were deemed to be locally-acquired. Biosurveillance findings highlight the potential for vector-borne cases among personnel at Joint Base San Antonio (JBSA), Texas 8% of the installation's military working dogs tested positive for T. will experience Chagasic heart disease and/or digestive megasyndromes.² Although immigrants from Latin America comprise the majority of cases in the United States, the cruzi antibodies in 2007.8 28% of 830 tratomine insects collected on the Medina Training Annex between 2013 and 2015 tested positive for T. cruzi parasites (personal communication, Walter Roachell, July 2015); and 33% of 131 insects from the area tested positive for human blood on the prevalence and seroprevalence of Chagas disease among personnel who may be at increased risk due to field with Trypanosoma cruzi, the causative parasitic agent of incidence of autochthonous disease may be increasing.3 Of blood meal analysis. This study was initiated to determine At least 300,000 U.S. residents are thought to be infected Chagas disease. 1 Approximately 30% of infected individuals activities conducted in T. cruzi-endemic areas.

Study Design Cross-sectional prevalence and seroprevalence study

Ouestionnaire

Participants were asked about their type and duration of field exposure, blood transfusion history, maternal living history, travel history, amount and type of outside exposure (camping, hunting), exposure to wildlife animals (raccoon, opossum, skunk, wood rats, deer, coyote, and feral hog), and history of bites by triatomines or unknown insects

Laboratory Tests

- RNA (18S rRNA gene). A sample was considered positive Real-time Polymerase Chain Reaction (PCR): whole cruzi parasite. Two multiplex TaqMan PCR assays were repetitive T. cruzi genomic regions: nuclear mini-satellite blood specimens were evaluated for presence of the T performed in parallel targeting highly conserved and TCZ). kinetoplast or kDNA, and small subunit ribosomal if all three targets were positive.
- Sera were serially diluted and placed on unbound serum antibodies, stained with a fluorescein Indirect Immuoflourescent Antibody (IFA): specimen sera antigen-coated microscope slides, washed to remove sothiocyanate-labeled anti-human IgG conjugate, and were evaluated for presence of human antibodies to T. visualized through a fluorescence microscope. A sample was considered positive for titers at or greater than 1:128. cruzi antigens.
- IgG conjugated to horse radish and washed again. HRP substrates Argentina) employs recombinant T. cruzi antigens to detect human IgG/IgM. Serum samples were incubated with the immobilized antigens, washed, incubated with peroxidase (HRP), and washed again. HRP substrates tetramethylbenzidine and hydrogen peroxide were then Chagatest recombinate v 3.0 assay (Wiener Laboratories added. The reactions were stopped with 2 N sulfuric acid color. The colorimetric readings were taken at 450 nm in a plate reader (ref wavelength, 650 nm). Cut-off, equivocal, reactive, and Enzyme-linked Immunosorbent Assay (ELISA): nonreactive values were defined as directed. resulted in yellowish

Oversight and Funding

This study was approved by the 59th Medical Wing Institutional Review Board, Funding was provided by the Air Force Medical Services Agency, Falls Church, Virginia, USA

N=416; April 1 - November 19, 2015 (enrollment ongoing) Enrollment and Surveillance Period Results

- USAF students graduating from security forces training at JBSA-Lackland and Camp Bullis
 - USAF instructors working at Basic Expeditionary Airman Skills Training at JBSA-Lackland, Medina Training Annex
 - Military working dog instructors at JBSA-Lackland

				les
Exposure in Weeks (average)	3,038 (7.3)	113,661 (273.2)	6.189 (14.9)	8,513 (20.5)
Table 1 Time in Triatomine-Endemic Environment	Field environment Lackland, AFB	Living/traveling Latin America or Southwest USA	Camping/hunting Latin America or Southwest USA	Wildlife exposure Latin America or Southwest USA

Personal and Family History [†]	Negative Pos	108
Received blood products in USA (N=314)	308	•
Received blood products outside USA (N=314)	312	
Mother lived in Latin America before birth (N=314)	288	2
Known triatomine bite (N=416)	416	Ü

Table 2

LaboratoryTest Results*	Negative	Positiv
PCR (N=403)	403	0
ELISA (N=408)	408	0
IFA (N=408)	408	0

1N<416 due to expanded questionnaire after initiation of study 1N<416 due to difficulty obtaining adequate blood aliquots on all enrollees</p>



Tristomine insect in a tent at Basic Expeditionary Airman Skills Training (courtesy of Walter Roachell, US Army Public Health Command-Central)



Adult females of the two most common Triatoma species collected on Joint Base San Antonio; gerstaeckeri (Jeft) and sanguisuga (right) (courtesy of Dr. Edward Wozniak, TX DSHS-Zoonosis Control)

Conclusions

- significant public health risk during military field exercises Chagas disease does not appear to constitute at JBSA-Lackland, Texas
- effectiveness of current measures (e.g., use of bed nets and insect repellent) to prevent triatomine bites and/or the poor efficacy of stercorarian transmission of 7. cruzili demonstrate The lack of positive cases may
- Since only 13.9% (n=416) of total participants have been enrolled, this research effort will continue to increase the statistical power of the results

- Bern C, Montgomery SP. An estimate of the burden of Chagas disease in the United Strates, CD: 2009; 483-242. Ress Jr. A, et al. Chagas disease Lancet, 2010; 375:1388-402. Kuehn BM. Putting Chagas disease on the US radar screen. JAMA.
 - 2015; 313(12):1195-7.
 McPhatter L, et al. Vector surveillance to determine species
- composition and occurrence of Toruzi infection at three milliany installations is Stan Anthonio, Tessa US Army Med Dept. 2012/12-31. Human Chapse Casses in Toras Available at http://www.dshs.stalle t...us/dcu/dcasset/chapset/data, accessed 7 Mart 16. Worwelfer P. at a The improbable transmission of Trypanosoma cruzi to human. PLoS Neigl Too Dis. 2013. 7(11):e2505. 2

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Acknowledgements

We thank Manuel Caballeto, Arlinda Halli, David Hill, Samantha Hune, Constance Kowat, Jamie Myers, Richard Mcintosh, Lois Robinson, and Michele Tavish at 59 MDW for their help drawing blood and conducting ELISA tests; Tyler Ito, Hillary Kamm, Angel Osuna, and Francisco Sanchez at USAPHC-Central for conducting IFA and PCR tests.

The voluntary, fully informed consent of subjects used in this research was abeliand as required by 32 CFR 219 and DOI) 321 602_AFI 40-402. The views expressed are those of the authors and do not reflect the official views or policy of the Department of Defense and its Components.



DEPARTMENT OF THE AIR FORCE AIR EDUCATION AND TRAINING COMMAND

13 April 2016

MEMORANDUM FOR 59 MDW
ATTN: CAPT MARY T. PAWLAK

FROM: 502 ISG/JA

SUBJECT: Ethics Review for Presentation Approval Request (Pawlak)

- 1. Capt Pawlak submitted a request for a legal review of a poster titled "Prevalence of *Trypanosoma cruzi* among U.S. Military Members Training in South Texas." The author plans to present this research as a poster at the following meetings: Diseases in Nature on 25-27 May 2016 in San Antonio, TX; Military Health System Research Symposium on 15-18 August 2016, in Kissimmee, FL; and American Society of Tropical Medicine and Hygiene on 13-17 November 2016 in Atlanta, GA. This legal review is limited to the ethics issues regarding the presentation. The abstract contains the required disclaimer discussed below. The disclaimer must also be included on the poster. The abstract must also be submitted for Public Affairs approval. There are no conflict of interest issues with presenting this research at these events.
- 2. FACTS: Capt Pawlak plans to present her poster titled "Prevalence of *Trypanosoma cruzi* among U.S. Military Members Training in South Texas" at the above listed events.
- 3. LAWS AND REGULATIONS: DoD 5500.07-R, Joint Ethics Regulation (JER), section 3-305 lays out rules governing "Teaching, Speaking and Writing." If the presentation will "deal in significant part with any ongoing or announced policy, program or operation" of the Air Force, the presenter is required to include a disclaimer that states the "views presented are those of the speaker or author and do not necessarily represent the views of DoD or its Components."
- 4. ANALYSIS: The presentation does not "deal in significant part with any ongoing or announced policy, program or operation" of the Air Force, however, the abstract does address research done as part of the author's military medical practice. Additionally, the presenter's affiliation with the military will be included as part of the presentation. The author has included the required disclaimer that the views presented are those of the authors and do not necessarily represent the views of DoD or its Components on the slide presentation. The disclaimer must be included on the poster. There are no apparent conflicts of interest that would prohibit the presentation.
- 5. CONCLUSIONS: The abstract presented for review included the disclaimer required by the JER. The disclaimer must be included on the poster.

6. If you have any questions, please call me at 210-671-5771.

VERNISHA N. FOSTER, Captain, USAF Assistant Staff Judge Advocate

e Christielen

l concur.

ARLENE R. CHRISTILLES

Chief, Civil Law